Groundwater
Monitoring&Remediation

Concentrations of Artificial Sweeteners and Their Ratios with Nutrients in Septic System Wastewater

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Abstract

This study reports the first comprehensive data set of characteristic concentrations of four artificial sweeteners: acesulfame (ACE), sucralose (SUC), saccharin (SAC), and cyclamate (CYC), and their ratios with nutrients, for untreated septic system wastewater. Samples were collected from the tanks of 19 different septic systems from across Ontario, Canada; these had a variety of usages, from single-family cottages to multiple-dwelling (campground or resort) facilities and had no additional treatment systems. The artificial sweetener concentrations and their relative proportions were highly variable in some cases, both temporally for several individual tanks and from site-to-site. Variability tended to be lower for multiple-dwelling compared to single-dwelling systems. This variability likely reflects differing use of artificial sweetener-containing products. The median concentrations for the complete data set of all four artificial sweeteners (in a range of 10 to 60 µg/L) were of a similar order of magnitude, but slightly higher, than has generally been reported for wastewater treatment plant influent (though these vary substantially globally). Both SUC and ACE provided adequate positive linear relationships for dissolved nitrogen and phosphorus in the septic tanks, while a summation of ACE and SUC concentrations also gave a strong correlation. In contrast, CYC and SAC showed poor linear correlation with these nutrients. These reported ranges for artificial sweetener concentrations and ratios with nutrients may be used in future studies to estimate the contributions of nutrients or other wastewater constituents (e.g., pharmaceuticals, bacteria, and viruses) from domestic septic systems to groundwater, including water supply or irrigation wells, and nearby surface water bodies.

Introduction

Artificial sweeteners such as acesulfame (ACE) and sucralose (SUC) have recently emerged as powerful tracers of wastewater in the environment (Buerge et al. 2009; Oppenheimer et al. 2011, 2012; Liu et al. 2014). They are widely used in various foods and beverages as well as in some pharmaceuticals and other household products (e.g., toothpaste) (see review by Lange et al. 2012). Current analytical methods can detect part-per-trillion levels of these compounds in groundwater and surface water samples (Buerge et al. 2009; Lange et al. 2012; Gan et al. 2013; Tran et al. 2014). Furthermore, both ACE and SUC are fairly resistant to attenuation and degradation in municipal wastewater treatment systems (Scheurer et al. 2009; Soh et al. 2011; Kokotou and Thomaidis 2013; Burke et al. 2014), rural septic systems (Robertson et al. 2013, 2016a, 2016b), and drinking water treatment plants (Mawhinney et al. 2011). Other sweeteners, such as saccharin (SAC) and cyclamate (CYC), are more readily degraded in wastewater treatment systems (Lange et al. 2012; Lim et al. 2017), but may still be useful wastewater tracers for nonquantitative detection and source identification (e.g., Van Stempvoort et al. 2013). And finally, given that there are no natural sources of these artificial sweeteners, background concentrations are usually below detection limits, which yields powerful resolution of even trace amounts of wastewater effluent. Thus, by measuring the levels of artificial sweeteners in environmental samples, the discharge of wastewater to groundwater, and potentially then to surface water systems can be identified and tracked, even with substantial dilution. However, in order to quantify effluent contributions and determine loadings of specific contaminant compounds, the concentrations of artificial sweeteners in wastewater sources must be well-characterized.

The range of concentrations of artificial sweeteners in untreated and treated wastewater at municipal wastewater treatment plants (WWTP) has been provided in several studies (see references in Table 1). However, to the best of our knowledge there is currently only one published report (Oppenheimer et al. 2011; only for SUC) that considers typical artificial sweetener concentrations in septic system wastewater, excluding the ACE data for several sites that are reported here for this study that were previously published by Robertson et al. (2016a). Wastewater from septic systems differs from that received by WWTP in that individual septic systems integrate waste from a much smaller number of people and will typically not have inputs from commercial or industrial activities or stormwater runoff. We report here a compilation of concentrations of four artificial sweeteners: ACE, SUC, SAC, and CYC, from several different septic systems across Ontario, Canada. The septic systems chosen for this study have varying usage; from small single-family

doi: 10.1111/gwmr.12229

Table 1

Summary Statistics of Artificial Sweetener Concentrations Found in Untreated Wastewater in Septic Tanks (Present Study and Others), Sewers, and WWTP Influent and Primary Lagoons

	Septic Tanks			Sewers	Lagoon	WWTP Influent						
	Single Dwelling (Present Study)	Multiple Dwelling (Present Study)	USA[1]	Germany[2]	Canada[3]	USA[4,5]	Europe[5,6]	Sweden[5,7,8]	Switzerland[5,9]	Germany[5,10,11]	Singapore[12]	Vietnam[13]
Sample count (*n*)	14	36	8	64	3	14	90	14	10	7	50	7
ACE (µg/L)												
Minimum	3.4	6.4			13	0.05	< 0.01		12	8	0.2	0.055
Maximum	190	160			19	2.3	2500		43	47	57	0.58
Median	57	32		42			14				12	0.36
SAC (µg/L)												
Minimum	6.1	2.2			9.7	1.1			3.9	34	1.5	7.6
Maximum	72	63			14	25			18	43	140	13
Median	22	16									22	13
CYC (µg/L)												
Minimum	0.003	0.003			12				10	141	1.4	0.087
Maximum	36	170			22				65	195	250	0.81
Median	2.0	14		250							17	0.58
SUC (µg/L)												
Minimum	2.2	5	12		35	16	>0.1	1.7	2	0.4	0.08	0.75
Maximum	360	110	80		43	46	13	7.9	9	1.5	5.2	1.5
Median	51	28					1.7				1.5	1.1

Raw data and additional statistics for this study are provided in Tables S1 and S2 in Appendix S1. A subset of the acesulfame data for multiple-dwelling sites (this study) were previously published by Robertson et al. (2016a).
[1]Oppenheimer et al. (2011); [2]Zirlewagen et al. (2016); [3]Van Stempvoort et al. (2011); [4]Subedi and Kannan (2014); [5]6- or 24-h composite samples; [6]Loos et al. (2013); [7]Brorström-Lundén et al. (2008); [8]Schmid Neset et al. (2010); [9]Buerge et al. (2009); [10]Scheurer et al. (2009); [11]Scheurer et al. (2011); [12]Tran et al. (2014); [13]Watanabe et al. (2016).

seasonal cottages to seasonal campground facilities and a resort, with no additional treatment systems (e.g., denitrifying filter). We also report the concentrations of chloride (Cl^-), and nitrogen (N), and phosphorus (P) nutrients. The temporal and intersite variability of the artificial sweetener concentrations and their ratios with nutrients is examined with the objective of establishing ranges appropriate for septic system wastewater. This information can then be used in future studies to estimate the contributions of nutrients or other wastewater constituents (e.g., pharmaceuticals, bacteria, and viruses) from septic system effluent to groundwater, including water supply or irrigation wells, and nearby surface water bodies. This in turn would provide guidance for the design and management of wastewater treatment systems to minimize adverse environmental and human health impacts.

Methods

Sampling and Analyses

Nineteen different septic systems from southern and central Ontario were sampled for this study, the details of which are outlined in Table 2, with some also described by Robertson et al. (2016a). All wastewater samples were obtained directly from within the septic holding tanks, using a peristaltic pump connected to ¼-in polyethylene tubing. A piece of nylon screen (100 µm) was secured to the opening of the tubing to filter out large solids in most cases. The samples were then filtered in the field using disposable 0.45-µm Whatman syringe filters (GE Healthcare Life Sciences, Mississauga, Ontario, Canada). Separate subsamples were collected in polyethylene bottles for artificial sweeteners, ammonium, major anions (including chloride, nitrate, and nitrite), and soluble reactive phosphorus (SRP) and/or total phosphorus (TP). The ammonium samples were immediately acidified to pH 5 to 6 with 10% hydrochloric acid; some SRP samples were also acidified with 10% hydrochloric acid to extend preservation time. Samples were kept cold during transport and then stored cold or frozen, depending on the subsample, until analysis.

Artificial sweetener samples were stored frozen prior to analysis by ion chromatography coupled to a tandem mass spectrometer with an electrospray ionization source in negative mode (Van Stempvoort et al. 2011) at Canada Centre for Inland Waters (CCIW; Burlington, Ontario, Canada). The detection limits for ACE, SAC, CYC, and SUC for years 2008 to 2012 were 8, 21, 3, and 5000 ng/L, respectively. System upgrades improved the detection limits to 2, 2, 3, and 20 ng/L, respectively, for samples collected in years 2013 to 2016. Major anions were analyzed at CCIW or at the University of Waterloo (UW) by ion chromatography, providing detection limits of <0.1 mg/L for each compound. Ammonium concentrations were determined at CCIW or UW by spectrophotometry (salicylate-nitroprusside colorimetric method) with absorbance measurements at 640 nm, providing a detection limit of <0.05 mg/L N. Measurements of SRP were made at CCIW or at the Soil and Nutrient Laboratory, University of Guelph (Guelph, Ontario, Canada) by spectrophotometry (orthophosphate method) with absorbance measurements at 885 nm, with a detection limit of 0.05 mg/L P. TP samples were analyzed at the University of Waterloo by spectrophotometry, with absorbance measurements at 885 nm, providing a detection limit of 0.05 mg/L. Samples were first digested with a saturated potassium persulfate reagent in a 100 °C bath for 1 h, before a mixed reagent consisting of ascorbic acid, de-ionized water, and a premix solution (ammonium molybdate, antimony potassium tartrate, and concentrated sulfuric acid) was added to each sample.

Table 2

Description of the 19 Sites for Septic Tank Sampling, with Number of Dates Sampled

Site(s)	Times Sampled (Years)	Site Details
C1 to C9	1 each (2011/2015)	Nine single-family cottages; seasonal to year-round access; some with dishwashers and laundry
C10	1 (2015)	Recreational lodge; occasional use, spring to fall, by maximum of 10 people
C11	1 (2013)	Two-person, permanent dwelling
C12	3 (2012)	Family cottage, regularly used May to November
P1	3 (2016)	Overnight campground with 157 campsites open April to October; flush toilets, showers, and laundry
P2	4 (2012)	Overnight campground with 118 campsites open May to October; flush toilets, showers, and laundry
P3	3 (2012)	Overnight campground with 128 campsites; three-season comfort station; flush toilets and showers
P4	10 (2010/2011)	Overnight campground with 253 campsites open May to October; flush toilets and showers
P5	4 (2015/2016)	Overnight campground with 144 campsites open May to October; flush toilets and showers
P6	6 (2015/2016)	Overnight campground with 231 campsites open May to October; flush toilets, showers, and laundry
LJ	5 (2012/2013)	Resort open May to September; average daily occupancy is 85 persons

Statistics

Prior to statistical analyses (determining mean, standard deviation, etc.), all data were assessed for normality using SOFA Statistics version 1.4.3 (Paton-Simpson & Associates Let., Auckland, New Zealand). The artificial sweeteners,

chloride, and TP data were not normally distributed; and the total inorganic nitrogen (TIN; sum of ammonium, nitrate, and nitrite) and SRP data passed conditionally (not considered 'perfectly' normal). Application of the Box-Cox transformation technique indicated a natural log transformation was the most appropriate (best transformation coefficient (λ) was zero; Figure S1a and S1b in Appendix S1, Supporting Infomation). After $\log_e$ transformations were complete, outlying data at the tails of the distributions were removed. In total, 16 data were removed from the SAC, CYC, and SUC datasets. With the exception of three high SAC values, all were low outliers at or near the practical quantification limits for the method.

Model II (major axis) regression (Legendre and Legendre 1998) was used to assess relationships between the different wastewater compounds measured. However, samples for both SRP and TP were not taken at all sites, with SRP being the most prevalent. Thus, to allow for a more complete statistical assessment of the phosphorus data, a Model II simple linear regression (major axis) was performed to determine if there was a relationship between TP and SRP data (Legendre and Legendre 1998). Four pairs of data (plus a datum at the origin) were regressed against each other, and the equation of the line had a slope of 0.704 ($R^2 = 0.956$), meaning approximately 70% of the TP value was comprised of SRP. This relationship was then used to gap-fill SRP where TP data existed but SRP data were missing, adding three more SRP data points.

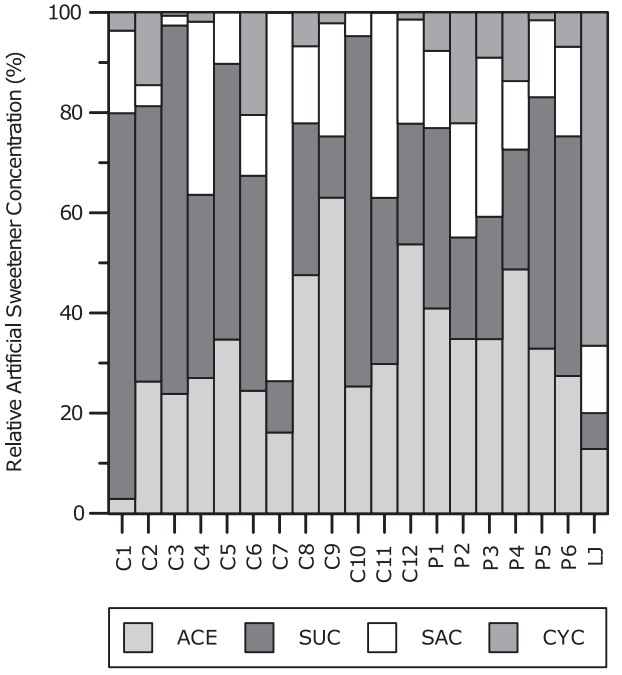

Figure 1. The relative concentrations (%) of ACE, SUC, SAC, and CYC in septic wastewater at each of the sites sampled in this study. Sites C1 to C11 were sampled once. The remaining sites were sampled multiple times (Table 2); mean values shown here were calculated from $\log_e$-transformed values and then converted using the anti-log(e^x), because these parameters were not normally distributed (Figure S1a in Appendix S1).

Results and Discussion

Artificial Sweetener Concentrations in Septic Tanks

All four artificial sweeteners were detected together at the majority of sites (site descriptions in Table 2), except for three sites with no CYC detected (Figure 1). ACE and SUC tended to be the dominant sweeteners (i.e., with the highest concentrations); however, SAC and CYC were each dominant at one site, a single cottage and resort, respectively. The range of concentrations (maximum to minimum; Table 1) covered about 2 orders of magnitude for ACE and SUC, but nearly 3 and >4 orders of magnitude for SAC and CYC, respectively. These results suggest more ubiquitous household use of the former two sweeteners for this study area, but generally widespread use of all four of the sweeteners.

All four sweeteners had similar median concentrations (generally between 10 and 60 µg/L, though higher for ACE and SUC, Table 1; mean concentrations were similar, Table S2 in Appendix S1), in both single- and multiple-dwelling septic tanks. The only other study (independent of this one) reporting artificial sweetener concentrations for septic tanks was for sites in the United States (Oppenheimer et al. 2011), though only SUC was measured and on fewer samples (Table 1). While the concentration ranges were greater for septic tanks of our study, for both single- and multiple-dwelling data sets, our median (and mean, Table S2 in Appendix S1) SUC concentrations fell within the range reported by Oppenheimer et al. (2011).

A previous study that looked at artificial sweeteners in wastewater in Canada sampled two wastewater lagoons in a different area of the country (Van Stempvoort et al. 2011) and found all four sweeteners at similar concentrations (typically ranging from 10 to 40 µg/L), with their maximum concentrations similar or higher than the median concentrations of the septic tanks in this study (Table 1). These two studies suggest that all four sweeteners are broadly used in Canada, though SUC appears to be dominant. In comparison, maximum concentrations in landfill leachate from Canada were 85, 250, and 14 µg/L for ACE, SAC, and CYC, respectively (Roy et al. 2014; SUC not assessed). Generally these are of a similar magnitude as found for the septic tanks in this study, though with a difference in the most dominant sweetener; for septic tank wastewater, the SAC maximum concentration (72 µg/L) was about half that of ACE and CYC (180 and 170 µg/L, respectively).

Many more studies, sampling from a variety of countries, have reported artificial sweetener concentrations for WWTP, often for both influent and effluent wastewaters. Here we consider the untreated WWTP influent wastewater most relevant for comparison to septic tank concentrations, because the former commonly involves treatments designed to promote microbiological reactions while septic tanks generally do not receive such treatments (though some microbiological activity presumably will occur naturally in the septic tanks). The range of concentrations for these same four sweeteners reported for WWTP influent (Table 1) typically ranged over several orders of magnitude in each study. While statistical comparisons between these studies were not performed, geography appears to be a key factor.

For example, SUC concentrations were notably higher in the WWTP influent from the United States, while that from Vietnam had much lower concentrations of all sweeteners but SAC. As noted by Lange et al. (2012), the presence of the different artificial sweeteners can vary substantially between countries due to differences in approval and usage. This likely is a key reason why the largest range of concentrations was typically from the Europe-wide study by Loos et al. (2013). Geographical variability complicates a comparison of this broader set of WWTP data to those for septic tanks. However, generally, the maximum concentrations observed in our study's septic tanks were higher than for the majority of the WWTP influents reported in other studies (Table 1; less so for CYC). Furthermore, we report a maximum concentration for SUC (360 µg/L) that is much higher than previously reported maximums for wastewater (80 µg/L for septic tanks in the United States; 46 µg/L for WWTP influent in the United States; Table 1). These differences may reflect disparities in sweetener use, with potentially greater use in Canada than in many of the other countries studied. Interestingly, a comparison across all of the studies (Table 1) does suggest that SUC use is more prevalent in North America, compared to Europe and Asia. Alternately, the septic – WWTP difference in concentrations may result from greater dilution of WWTP influent with nonwaste sources (e.g., industrial waters, stormwater runoff) that do not influence domestic septic systems.

In addition, the minimum concentrations of CYC were an order of magnitude lower for this study's septic systems than have been reported for WWTP influent (Table 1). This likely reflects the lower number of contributors to septic tanks (i.e., one or several families only in some cases; Table 2) compared to WWTPs, combined with less ubiquitous use of this sweetener in the study area (cottage area in Canada). It is also possible that biodegradation of CYC in the septic tanks may also play a role here; further study would be needed to address this.

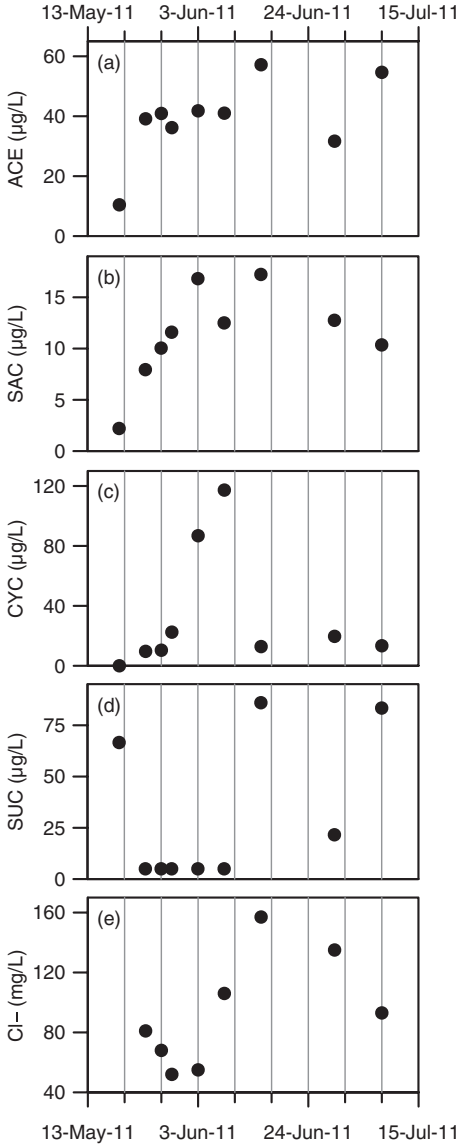

Figure 2. The temporal variation in septic tank concentrations of (a) ACE, (b) SUC, (c) SAC, (d) CYC, and (e) Cl^- at site P4 over a 50-d period (*x*-axis tick marks represent 1 week).

Temporal Variability

A temporal series of septic tank samples was collected at all of the multiple-dwelling sites and one of the single-dwelling sites (C12). Data from site P4, a three-season campground, are shown in Figure 2 to illustrate the substantial changes that can occur in sweetener concentrations and in their ratio to each other, even over short time intervals (i.e., several days). In the P4 tank, CYC showed the greatest range in concentration, with a relatively sharp peak in early June, while ACE and SAC were less variable (except for the first data point). During the first month of sampling, SUC showed a reverse pattern to that of ACE, but the patterns were similar in the second month. These findings illustrate that the temporal variation in individual sweetener concentrations within septic wastewater can be substantial and complex. The remainder of sites generally showed less extensive temporal changes for sweetener concentrations compared to P4 (Appendix S1).

Changes in chloride concentration may indicate variability in the contribution from other water sources (e.g., showers, sinks, etc.) to the septic tank wastewater, because chloride concentrations are less likely to vary for human waste compared to the individual artificial sweeteners. The chloride concentration for the septic tank at site P4 ranged by a factor of 4 over the approximate 2-month sampling period, generally increasing to higher values in the early summer (Figure 2). In comparison, the sweetener concentrations all ranged by an order of magnitude, and none of these mirrored the temporal changes in Cl^- concentration (with SUC most similar). This indicates that dilution (e.g., from showers and sinks) was not the primary driver of the temporal variation observed for the sweetener concentrations.

Another possible cause of the variation in sweetener concentrations would be changes in the rates of biodegradation, which would be most likely for SAC and CYC. For instance, higher temperatures in the summer (assumed, not measured) may have boosted microbial degradation rates of CYC in the septic tank, possibly contributing to the summer

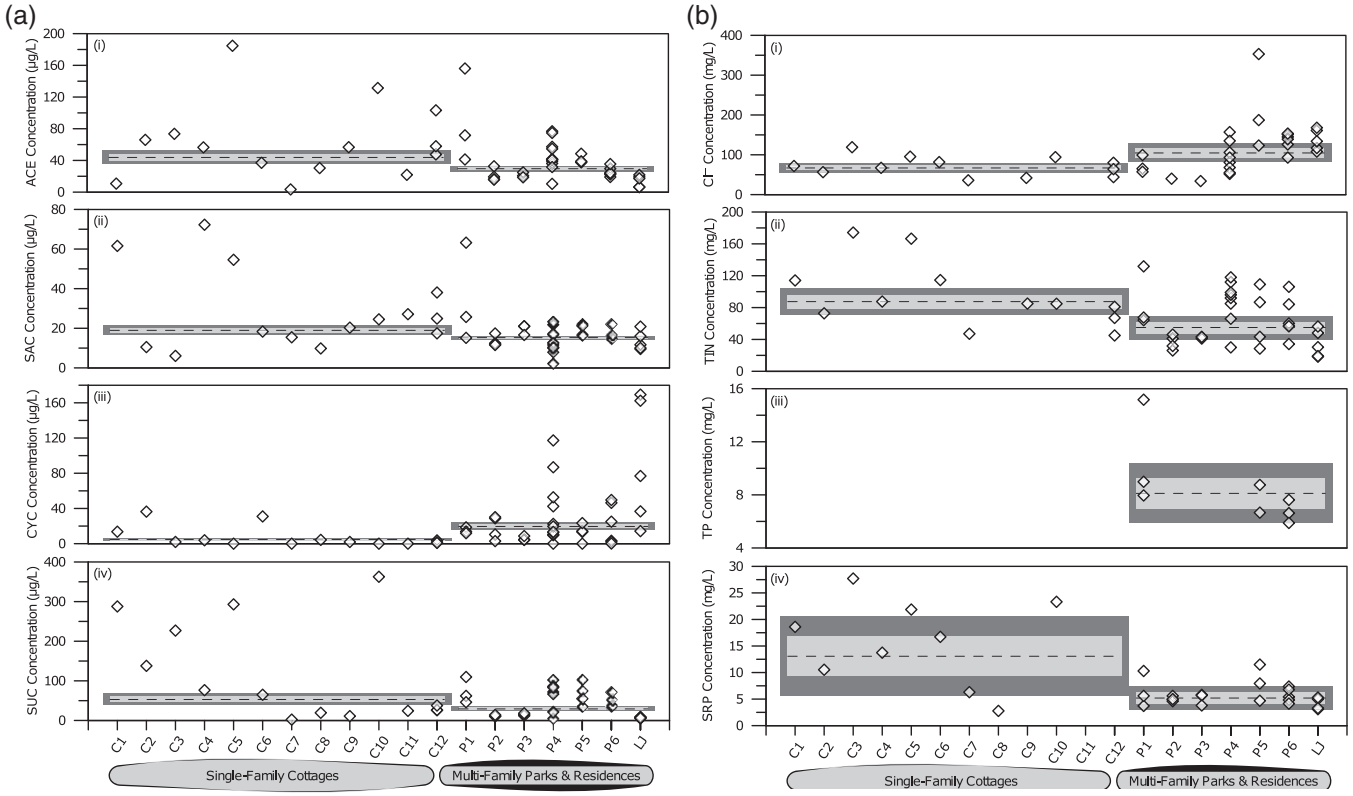

Figure 3. The intersite variations of (a) artificial sweetener concentrations in septic tanks at single-family cottages and multi-family parks and residences; and (b) Cl^-, TIN, TP, and SRP concentrations in septic tanks at single-family cottages and multi-family parks and residences. The mean values (dashed lines) are encompassed by boxes representing 1 standard deviation from the mean (light gray box) and 2 standard deviations from the mean (dark gray box). All data are shown (diamond symbols), but see *Methods* section for calculation description for mean and standard deviations.

decline in CYC concentrations (Figure 2). However, we suspect that the most important cause of the temporal variations in sweetener concentrations, being independent of Cl^- and differing by type of sweetener, was changes in the sweetener loadings to the septic tank (though we have no direct proof of this). This would reflect differences in the food and drink ingested and/or in the use of sweetener-containing products by the varying clientele at the P4 campground. Thus, we suspect that temporal variation in sweetener concentrations could be even more extreme for single-dwelling cottages with changing occupants, such as those that are rented out for substantial periods or shared across an extended family.

Variability across Sites

There was substantial variation in septic tank sweetener composition between sites, both in terms of the magnitude of concentrations (Figure 3a) and the relative proportion of the four sweeteners (Figure 1). This was found even for the two dominant sweeteners, as ACE and SUC made up between 5% (ACE) or 10% (SUC) and 75% of the total sweeteners (Figure 1), though typically both ACE and SUC were in the range of 20 to 50%. The variation in sweetener concentration (Figure 3a) was not consistent across the four sweeteners. For some sites, several sweeteners would have elevated concentrations (e.g., C5 and C10), but at others it was only one sweetener that was notably high (e.g., SUC at C1 and CYC at LJ). This likely reflects differences in the use of sweetener-containing products (e.g., brand of coffee sweetener) between users of the different sites.

For ACE, SAC, and SUC, there was less variation in concentrations (closer to mean values) for multiple-dwelling sites compared to single-dwelling sites (Figure 3a), as might be expected when there is a larger pool of waste contributors, presumably utilizing a wider variety of sweetener-containing products. The variation in CYC concentration was the opposite. The reason why several multiple-dwelling sites had such high CYC concentrations (Figure 3), in contrast to the majority of single- and multiple-dwelling sites, is not known (though it is possible that washroom disinfection activities added some chemical to these tanks that inhibited CYC biodegradation). This suggests that studies involving a low number of septic systems will be more prone to uncertainty if average values for sweetener concentrations from this study are applied. However, such uncertainty will likely diminish for higher numbers of septic systems or for larger systems with more contributors.

Relationship with Other Wastewater Compounds

Concentrations of other wastewater constituents, Cl^- and nutrients (TIN, SRP, and TP), varied within an order of magnitude across all of the studied septic tanks (Figure 3b); which is less than was found for the four sweeteners (Figure 3a), as described above. Higher Cl^- concentrations at a few of the multiple-dwelling sites (especially P4) are

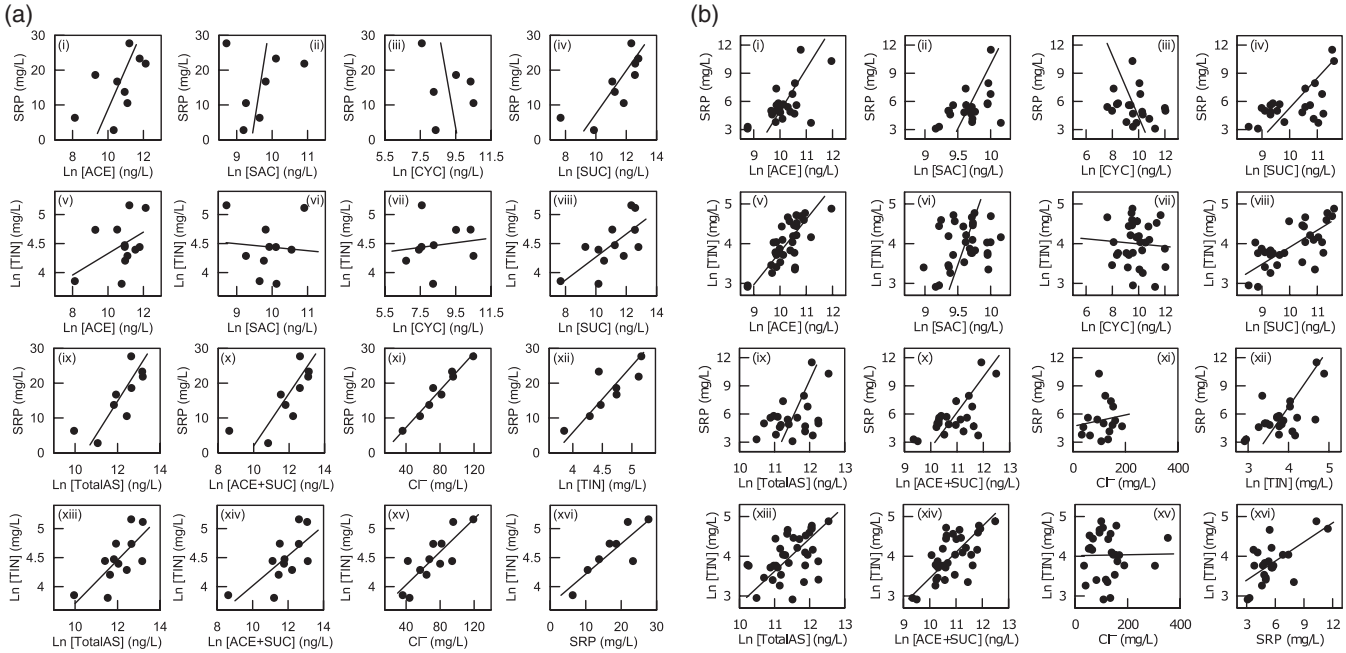

Figure 4. Linear correlations between (a) artificial sweeteners, Cl^-, TIN, and SRP at single-family cottages (sites C1 to C12); and (b) artificial sweeteners, Cl^-, TIN, and SRP at multi-family parks and residences (sites P1 to P6, LJ). Concentrations of ACE, SAC, CYC, SUC, total artificial sweeteners (TotalAS), ACE + SUC, and TIN were log_e-transformed to improve normality and the goodness of linear fit, which was evaluated by the coefficient of determination (r^2). The Major Axis (Model II) linear regression line is shown for each correlation, and the equation parameters are listed in Table S3 in Appendix S1.

likely associated with the use of salt in water softeners, which raised the average Cl^- concentration for the multiple-dwelling sites. Both TIN and SRP were higher on average for single-dwelling sites compared to multiple-dwelling sites; the cause may be less dilution from nonwaste waters for single-dwelling sites (i.e., less shower and laundry use perhaps). This reduced variability bodes well for applying these average values to nutrient loading models where chemical analyses of septic system effluents are not readily available.

Linear correlations of the artificial sweeteners and inorganic constituents were performed separately for the single-dwelling (Figure 4a) and multiple-dwelling (Figure 4b) sites (Table S3 in Appendix S1 provides regression parameters). There was a relatively strong correlation between SRP (with a few points converted from TP) and TIN, with a better linear relationship for single-dwelling ($r^2 = 0.72$) than multiple-dwelling ($r^2 = 0.39$) sites. The best correlation existed between Cl^- and SRP at the single-dwelling sites ($r^2 = 0.95$), suggesting consistent concentrations of these constituents in septic wastewater at single-dwelling sites. This relationship was poor at the multiple-dwelling sites ($r^2 = 0.02$), as Cl^- values at some sites were likely elevated by salt used for water softening. Thus, in areas dominated by single-dwelling cottages, Cl^- would be a strong candidate as a predictor for wastewater inputs of dissolved P. However, once the wastewater was discharged to the environment, other inputs of Cl^- such as from road salt or natural rock weathering would act to interfere with this relationship. These confounding sources are the reason that wastewater-specific compounds, such as artificial sweeteners, have been sought to replace the use of Cl^- as a wastewater tracer.

The relationships between the four individual sweeteners and the dissolved nutrient species SRP and TIN were not as strong (Figure 4a and 4b; Table S3 in Appendix S1). The best relationships for SRP and TIN for (1) single-dwelling sites was with SUC ($r^2 = 0.62$ and 0.48, respectively; Figure 4a), and (2) for multiple-dwelling sites was ACE ($r^2 = 0.39$ and 0.55, respectively; Figure 4b). The relationships were generally of similar quality when the concentrations of ACE and SUC were added, or the sum of all four sweetener concentrations was used. Meanwhile, SAC and CYC relationships with the other wastewater parameters were poor for both data sets, which is not surprising considering the larger variation found for the SAC and CYC concentrations (Table 1) and their greater propensity for undergoing biodegradation (Lange et al. 2012). These findings suggest that both SUC and ACE would make suitable tracers for SRP and TIN sourced from septic systems in the study area and areas with similar sweetener usage. Performing parallel calculations with both sweeteners, if available, as well as the combined concentration of ACE and SUC, would likely provide greater confidence in the results. The use of SAC and CYC for quantifying wastewater inputs from septic systems is not recommended based on these results.

Conclusions

This study reports the first comprehensive data set for the concentrations of four artificial sweeteners, ACE, SAC, CYC, and SUC in untreated wastewater of domestic septic systems. The results may not apply where secondary systems (e.g., denitrifying filter) are used, because these may

lead to enhanced degradation of some of these artificial sweeteners as has been reported for WWTP effluent (e.g., Lange et al. 2012). The resultant median concentrations (generally falling in a range of 10 to 60 µg/L for all four artificial sweeteners) may be used to represent concentrations in septic tanks where direct measurements are lacking, in modeling or mass balance calculations of septic wastewater environmental loadings in areas with similar usage of these artificial sweeteners. For example, Robertson et al. (2016a) and Spoelstra et al. (unpublished data) used ACE concentrations in groundwater samples (wells, streambed profiler, and/or seeps) to estimate the fraction of water derived from septic system wastewater. As such, artificial sweetener analysis of water samples gives insight into potential health risks due to the possible presence of other wastewater constituents such as nitrate, bacteria and viruses, and pharmaceuticals. Such calculations could also be incorporated into lake-wide nutrient balance studies to evaluate the relative contribution of septic systems to phosphorus loading, a key driver of eutrophication in freshwater systems (Correll 1998). This could inform, for example, the review of standards for on-site septic systems and planning of future residential development in these areas.

The median sweetener concentrations for septic wastewater in this study were of a similar order of magnitude, but slightly higher, than has generally been reported for WWTP influent (though these show a very wide range globally). However, a general comparison of artificial sweetener concentrations of WWTP influent across many different countries suggests that these septic tank concentrations may not be representative globally (due to differences in product restrictions and generally consumption patterns). Therefore, their application to countries besides Canada should be performed carefully, with guidance afforded by any available data of sweetener concentrations in any type of wastewater (septic, sewer, WWTP), or even landfill leachate. Alternately, direct measurement of these sweetener concentrations in septic wastewater could be made in localities where sweetener usage is expected to differ.

The study findings also demonstrate that the sweetener concentrations can be highly variable, both temporally in individual septic tanks and from site-to-site. Variability tended to be lower for those tanks receiving wastewater from multiple groups of people (i.e., families), as for septic servicing of campgrounds or apartment buildings, with similar or slightly lower median concentrations. Inconsistent ratios of the artificial sweeteners with chloride (Cl^-) suggest that this variability is likely due to variable use of products with artificial sweeteners, rather than a result of dilution from nonwaste water inputs.

Relationships for the concentrations of wastewater nutrients (dissolved N and P) in the septic tanks showed that Cl^- generally provided the strongest positive linear relationship for the pooled set of single-cottage sites where water softening treatment is not employed. However, the relationship with Cl^- was poor if sites with suspected water softening were included. Chloride also has many nonwastewater sources (e.g., road salt; natural groundwater) that may interfere with tracing wastewater effluent from septic systems, whereas this is much less of a problem for artificial sweeteners. Of the artificial sweeteners assessed, both SUC and ACE provided adequate positive linear relationships for dissolved N and P, while a summation of ACE and SUC concentrations also gave a strong correlation. These two sweeteners also show less susceptibility to biodegradation in the natural environment, though they may degrade in certain conditions (ACE – Castronovo et al. 2017 and references within; SUC - Robertson et al. 2016b); thus, these would provide the best tracers for quantifying potential loadings of septic system nutrients to surface waters, noting that nutrient uptake and transformation processes may occur that do not affect the sweeteners. Use of CYC and SAC is not recommended for this purpose, given their poor linear correlation with the nutrients (Figure 4a and 4b) and their greater propensity to biodegrade (Lange et al. 2012). This potential nutrient contribution from septic wastewater determined from ACE and/or SUC, could then provide guidance for policy and management measures that address eutrophication of freshwater systems.

Acknowledgments

Funding for this work was provided by the Lake Simcoe-Georgian Bay Clean-Up Fund of Environment and Climate Change Canada (ECCC). Sue Brown, Jacob Burbank, Pam Collins, and John Voralek (ECCC), and Jennifer Hickman, Wesley Palmer, and Kelita Pratt (U.Waterloo), provided lab and field assistance. Helpful comments from two anonymous reviewers are also greatly appreciated.

Supporting Information

The following supporting information is available for this article:

Appendix S1. Artificial sweeteners in untreated septic system wastewater.

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